



Patent Attorney's Docket No. <u>0001-00001CON1</u>

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IN THE UNITED STATES PATENT AND TRADEMARK OFFICE BEFORE THE BOARD OF PATENT APPEALS AND INTERFERENCES

In re Patent Application of)
Patricia CRUZ-PEREZ et al.) Group Art Unit: 1637
Application No.: 10/804,339) Examiner: S. Woolwine
Filed: March 19, 2004)
For: METHOD FOR DETECTION OF Stachybotrys chartarum IN PURE CULTURE AND FIELD SAMPLES USING QUANTITATIVE POLYMERASE CHAIN REACTION))))

APPEAL BRIEF

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Sir:

This appeal is from the decision of the Primary Examiner dated August 14, 2006 and in support of the Notice of Appeal filed November 14, 2006.

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I. REAL PARTY IN INTEREST

The real party in interest in this appeal is the University of Nevada, Las Vegas.

II. RELATED APPEALS, INTERFERENCES, AND JUDICIAL PROCEEDINGS

To the best of the knowledge of the undersigned, there are no other appeals, interferences or judicial proceedings known to the Appellants, the Appellants' legal representative, or the above-noted assignee that will directly affect or be directly affected by, or have a bearing on, the Board's decision in this appeal.

III. STATUS OF CLAIMS

Claims 18-27 are currently pending in the application. Claims 1-17 have been canceled. No claims have been allowed. Claims 18-27 were finally rejected in the final Office Action dated August 14, 2006.

IV. STATUS OF AMENDMENTS

Appellants have filed no amendments subsequent to the final Office Action dated August 14, 2006.

V. SUMMARY OF CLAIMED SUBJECT MATTER

In the paragraphs that follow, each of the independent claims that is involved in this appeal that is argued separately will be recited followed in parenthesis by examples of where support can be found in the specification and drawings.

Claim 18 recites a method for identifying and quantifying the presence of the fungus
Stachybotrys chartarum in a collected sample that includes obtaining a primer set and probe that
is specific for the fungal species Stachybotrys chartarum (paragraph [0037]) and collecting the
sample from the environment (step 405, FIG. 4; paragraph [0032]). The method further includes
extracting the sample's DNA (steps 410-530, FIGS. 4-5; paragraphs [0033] and [0034]),
obtaining DNA standards from a culture of Stachybotrys chartarum (steps 105-225, FIGS. 1-2;
paragraphs [0026] and [0027]) and determining the concentration of Stachybotrys chartarum
spores in the DNA standards (step 320, FIG. 3; paragraph [0029]). The method also includes
amplifying by polymerase chain reaction each of the DNA standards and the collected sample's
DNA using the obtained primer set and probe (step 710, FIG. 7; paragraph [0037]) and
comparing amplification plots obtained by polymerase chain reaction of each of the DNA
standards and the collected sample's DNA to obtain an indication of the presence of the fungus
Stachybotrys chartarum in the collected sample and a concentration of the fungus Stachybotrys
chartarum in the collected sample (steps 715 and 720, FIG. 7; paragraph [0038]).

Claim 19 further recites wherein the wherein the primer set and probe of claim 18 includes a forward primer comprising a base sequence (SEQ ID NO: 1)

5'GTTGCTTCGGCGGGAAC3', a reverse primer comprising a base sequence (SEQ ID NO: 2) 5'TTTGCGTTTGCCACTCAGAG3', and a probe comprising a base sequence (SEQ ID NO: 5) 6-FAM-5'CTGCGCCCGGATCCAGGC3'-TAMRA, wherein the forward primer, reverse primer and probe do not cross-react with other fungal species when used in combination in polymerase chain reaction (paragraph [0037]).

Claim 20 further recites wherein the primer set and probe of claim 18 includes a forward primer comprising a base sequence (SEQ ID NO: 3)

5'ACCTATCGTTGCTTCGGCG3', a reverse primer comprising a base sequence (SEQ ID NO: 4) 5'GCGTTTGCCACTCAGAGAATACT3', and a probe comprising a base sequence (SEQ ID NO 5) 6-FAM-5'CTGCGCCCGGATCCAGGC3'-TAMRA, wherein the forward primer, reverse primer and probe do not cross-react with other fungal species when used in combination in polymerase chain reaction (paragraph [0037]).

Claim 22 recites a method for identifying and quantifying the presence of the fungus *Stachybotrys chartarum* in a collected sample that includes obtaining a primer set and probe that is specific for the fungal species *Stachybotrys chartarum*, wherein the obtained primer set and probe comprises: a forward primer comprising a base sequence (SEQ ID NO: 1) 5'GTTGCTTCGGCGGGAAC3', a reverse primer comprising a base sequence (SEQ ID NO: 2) 5'TTTGCGTTTGCCACTCAGAG3', and a probe comprising a base sequence (SEQ ID NO: 5) 6-FAM-5'CTGCGCCCGGATCCAGGC3'-TAMRA (paragraph [0037]). The method further includes employing quantitative polymerase chain reaction, using the obtained primer and probe

set, to determine a concentration of the fungus *Stachybotrys chartarum* in the collected sample, wherein the forward primer, reverse primer and probe do not cross-react with other fungal species when used in combination in the quantitative polymerase chain reaction (steps 710-720, FIG. 7; paragraphs [0037] and [0038]).

Claim 25 recites a method for identifying and quantifying the presence of the fungus *Stachybotrys chartarum* in a collected sample that includes obtaining a primer set and probe that is specific for the fungal species *Stachybotrys chartarum*, wherein the obtained primer set and probe comprises: a forward primer comprising a base sequence (SEQ ID NO: 3) 5'ACCTATCGTTGCTTCGGCG3', a reverse primer comprising a base sequence (SEQ ID NO: 4) 5'GCGTTTGCCACTCAGAGAATACT3', and a probe comprising a base sequence (SEQ ID NO: 5) 6-FAM-5'CTGCGCCCGGATCCAGGC3'-TAM (paragraph [0037]). The method further includes employing quantitative polymerase chain reaction, using the obtained primer and probe set, to determine a concentration of the fungus *Stachybotrys chartarum* in the collected sample, wherein the forward primer, reverse primer and probe do not cross-react with other fungal species when used in combination in the quantitative polymerase chain reaction (steps 710-720, FIG. 7; paragraphs [0037] and [0038]).

VI. GROUNDS OF REJECTION TO BE REVIEWED ON APPEAL

Claims 18 and 21 stand rejected under 35 U.S.C. §102(b) as allegedly being anticipated by "Quantitative Measurement of *Stachybotrys chartarum* conidia Using Real Time Detection

of PCR Products with the TaqManTM Fluorogenic Probe System," R.A. Haugland et al., Molecular and Cellular Probes (1999) 13: 329-340 (hereinafter "HAUGLAND").

Claims 19, 20 and 22-27 stand rejected under 35 U.S.C. §103(a) as allegedly being unpatentable over HAUGLAND in view of "Design Strategies and Performance of Custom DNA Sequencing Primers," Bio Techniques 27:528-536 (September 1999) (hereinafter "BUCK") and GenBank GI: 3420911 (hereinafter "GENBANK").

VII. ARGUMENT

A. The rejection of claims 18 and 21 under 35 U.S.C. §102(b) as allegedly being anticipated by HAUGLAND should be reversed.

Independent claim 18 recites a method for identifying and quantifying the presence of the fungus *Stachybotrys chartarum* in a collected sample that includes obtaining a primer set and probe that is specific for the fungal species *Stachybotrys chartarum*, collecting the sample from the environment, extracting the sample's DNA, obtaining DNA standards from a culture of *Stachybotrys chartarum*, determining the concentration of *Stachybotrys chartarum* spores in the DNA standards, amplifying by polymerase chain reaction each of the DNA standards and the collected sample's DNA using the obtained primer set and probe, and comparing amplification plots obtained by polymerase chain reaction of each of the DNA standards and the collected sample's DNA to obtain an indication of the presence of the fungus *Stachybotrys chartarum* in

the collected sample and a concentration of the fungus *Stachybotrys chartarum* in the collected sample.

A proper rejection under 35 U.S.C. §102 requires that a reference teach every aspect of the claimed invention. See M.P.E.P. § 2131. HAUGLAND does not disclose or suggest each and every feature recited in Appellants' claim 18. For example, HAUGLAND does not disclose or suggest, among other features, "obtaining a primer set and probe that is specific for the fungal species *Stachybotrys chartarum*," or "comparing amplification plots obtained by polymerase chain reaction of each of the DNA standards and the collected sample's DNA to obtain an indication of the presence of the fungus *Stachybotrys chartarum* in the collected sample and a concentration of the fungus *Stachybotrys chartarum* in the collected sample," as recited in claim 18.

With respect to the feature "obtaining a primer set and probe that is specific for the fungal species *Stachybotrys chartarum*," the final Office Action (pg. 2) relies on the first sentence of the "Results" section on pg. 334 of HAUGLAND for allegedly disclosing this feature. Appellants submit that this section of HAUGLAND does not disclose or suggest a primer set and probe that is <u>specific</u> for the fungal species *Stachybotrys chartarum*, as recited in claim 18. This section of HAUGLAND discloses:

The sequences and target sites of the forward (StacF4) and reverse (StacR5) PCR primers and TaqMan probe (StacP2) constructed for the detection of *S. chartarum* rDNA sequences in this study are shown in Fig. 1.

It is further apparent from FIG. 1 of HAUGLAND (pg. 335) that HAUGLAND's forward primer (StacF4) is completely homologous to the rDNA sequences of *S. dichroa*, *M. echinata* and *M. subsimplex*. In addition, HAUGLAND discloses on pg. 334, column 1, second paragraph, that "Priming efficiency analysis indicated that the rDNA sequences of two of these species, *S. cylindrospora* and *S. oenanthes*, had the potential to be amplified by both StacF4 and StacR5." Thus, Appellants submit that the forward (StacF4) and reverse (StacR5) primers disclosed by HAUGLAND may amplify fungal species other than *S. chartarum* and, therefore, these primers are not "specific for the fungal species *Stachybotrys chartarum*," as recited in claim 18.

With respect to the feature "comparing amplification plots obtained by polymerase chain reaction of each of the DNA standards and the collected sample's DNA to obtain an indication of the presence of the fungus *Stachybotrys chartarum* in the collected sample and a concentration of the fungus *Stachybotrys chartarum* in the collected sample," the final Office Action (pgs. 3-4) relies on the last paragraph of the "Quantification of *S. chartarum* conidia using the comparative C_T method" section on pg. 332 of HAUGLAND for allegedly disclosing this feature. Appellants submit that this section of HAUGLAND does not disclose or suggest comparing amplification plots obtained by polymerase chain reaction of each of the DNA standards and the collected sample's DNA, as recited in claim 18. This section of HAUGLAND discloses:

Each series of DNA extracts was also analysed using only S. chartarum target sequence assay results. In these calculations, calibrator sample C_T values were subtracted directly from corresponding test sample C_T values to obtain $\Delta C_{T,STAC}$ values. These values were used in place of $\Delta\Delta C_T$ values to determine the ratio of target sequences in the test and calibrator samples and to quantify S. chartarum conidia in the test samples as described above.

Furthermore, on page 332, in the last sentence of the second paragraph of the "Quantification of S. chartarum conidia using the comparative C_T method" section, HAUGLAND discloses the use of "ratios of target sequences determined in the test and calibrator samples...to obtain estimates of the absolute quantities of these conidia in the test samples." Thus, HAUGLAND merely discloses the use of a mathematical formula for determining the ratio of the target sequences in test and calibrator samples. HAUGLAND does not disclose, suggest, or even mention, the comparison of amplification plots obtained by polymerase chain reaction to obtain an indication of the presence and concentration of the fungus Stachybotrys chartarum in a test sample. HAUGLAND, therefore, does not disclose or suggest "comparing amplification plots obtained by polymerase chain reaction of each of the DNA standards and the collected sample's DNA to obtain an indication of the presence of the fungus Stachybotrys chartarum in the collected sample," as recited in claim 18.

Since HAUGLAND does not disclose or suggest each and every feature of claim 18, HAUGLAND cannot anticipate claim 18. Reversal of the rejection of claim 18 under 35 U.S.C. § 102 is, therefore, respectfully requested.

Claim 21 depends from claim 18. Reversal of the rejection of claim 21 is, therefore, requested for at least the reasons set forth above with respect to claim 18.

B. The rejection of claims 19, 20 and 22-27 under 35 U.S.C. §103(a) as allegedly being unpatentable over HAUGLAND in view of BUCK and GENBANK should be reversed.

In rejecting claims 19, 20 and 22-27, the final Office Action asserts (pg. 9) that "[t]he only limitations...not taught by Haugland are the specific primers/probes (SEQ ID NOs 1-5) used for the quantification of *Stachybotrys chartarum*." The final Office Action further asserts (pg. 9) that "SEQ ID NOs 1-5 were all well known sequences of the 18S ribosomal RNA gene of Stachybotrys chartarum at the time the invention of the instant application was made as shown by GenBank GI:3420911." The final Office Action (pg. 11) additionally alleges that Appellants' claimed primers/probes "simply represent functional homologues of the primers/probes taught by Haugland" and, therefore, that "the claimed primers/probes are *prima facie* obvious over Haugland's primers/probes...." The final Office Action (pgs. 11-12) further cites to the disclosure of BUCK as allegedly providing evidence that Appellants' primers are obvious.

close structural similarities and similar utilities. In re Payne, 606 F.2d 303, 313 (CCP 1979).

See M.P.E.P. §2144.09. HAUGLAND discloses (pg. 335) a forward primer 5'
TCCCAAACCCTTATGTGAACC-3' and a reverse primer 5'
GTTTGCCACTCAGAGAATACTGAAA-3' for detecting Stachybotrys chartarum using polymerase chain reaction. In contrast, the present application claims forward primers (SEQ ID NO: 1) 5'GTTGCTTCGGCGGGAAC3' and (SEQ ID NO: 3)

A prima facie case of obviousness may be made when chemical compounds have very

5'ACCTATCGTTGCTTCGGCG3' and reverse primers (SEQ ID NO: 2)

5'TTTGCGTTTGCCACTCAGAG3' and (SEQ ID NO: 4)

5'GCGTTTGCCACTCAGAGAATACT3'. As can be seen by a comparison of the forward primer of the present application with the forward primer of HAUGLAND, the forward primer of HAUGLAND includes a different sequence of bases than the forward primers of the present application and, therefore, is not structurally similar. As can further be seen by a comparison of the reverse primers of the present application with the reverse primer of HAUGLAND, the reverse primer of HAUGLAND includes a different sequence of bases than the reverse primers of the present application and, therefore, is not structurally similar. Since the primers of the present application are not structurally similar to the primers of HAUGLAND, Appellants' submit that the primers disclosed in HAUGLAND do not establish a *prima facie* case of obviousness by themselves.

In the "Response to Arguments" section (pg. 13), the final Office Action newly asserts that the primers of HAUGLAND, and the primers of the Appellants' claimed methods, have "'very close structural similarities' in that both comprise an identical sugar-phosphate backbone which makes up a significant portion of the molecule." Appellants traverse this assertion. Primers are defined as "a short single-stranded DNA fragment that is required to initiate polymerization of new DNA nucleotides." (Morris, C. 1992. *In* Dictionary of Science and Technology, p. 1723. Academic Press, San Diego, CA). Furthermore, DNA is defined as "a polymer of nucleotides connected via a phosphate-deoxyribose sugar backbone." (Brock, T.D.,

M.T. Madigan, J.M. Martinko, and J. Parker. 1994. *In* Biology of Microorganisms, 7th Ed., p.870. Prentice Hall, NJ). Therefore, all primers by definition are composed of a sugarphosphate backbone. Based on the above-noted assertion of the final Office Action, any primer (since every primer has a sugar-phosphate backbone) would have "close structural similarity" to the primers of HAUGLAND regardless of whether that primer is a primer for detecting *S. chartarum* or for detecting <u>any other</u> living organism. Appellants submit that a primary structural difference for distinguishing primers and probes is the sequence of bases that comprise the primers and probes. As already discussed above, a comparison of the forward and reverse primers of the present application with the forward and reverse primers of HAUGLAND evidences that the forward and reverse primers of the present application and, thus, are not structurally similar, contrary to the new assertions of the final Office Action.

On page 11, the final Office Action cites to In re Deuel and alleges that "since the claimed primers/probes simply represent functional homologues of the primers/probes taught by Haugland, the claimed primers/probes are prima facie obvious over Haugland's primers/probes..." (emphasis added). From this allegation, it appears that the final Office Action is alleging that a prima facie case of obviousness is established because the primers and probe of HAUGLAND perform a similar function (e.g., detection of Stachybotrys chartarum) to the primers and probes of the present application, even though the primers of HAUGLAND are entirely different from, and structurally distinct from, the primers of the present application.

APPellants respectfully submit that the case law does not support the final Office Action's allegation. As noted above, a prima facie case of obviousness may be made when chemical compounds have very close structural similarities. See M.P.E.P. §2144.09. The case law does not support the proposition alleged by the final Office Action that chemical compounds that may perform similar functions are prima facie obvious over one another. The case cited by the final Office Action, In re Deuel, merely stands for the proposition that the existence of a general method of isolating DNA molecules is irrelevant to the question of whether the specific molecules would have been obvious in the absence of other prior art that suggests the claimed DNAs. See M.P.E.P. 2144.09. In re Deuel has nothing to do with "functional homologs" as alleged by the final Office Action and this is apparent from the portion of In re Deuel cited in the final Office Action (pg. 11):

Normally, a prima facie case of obviousness is based upon *structural similarity*, i.e., an established structural relationship between a prior art compound and the claimed compound. *Structural relationships* may provide the requisite motivation or suggestion to modify known compounds to obtain new compounds. For example, a prior art compound may suggest its homologs because homologs often have similar properties and therefore, chemists of ordinary skill would ordinarily contemplate making them to try to obtain compounds with improved properties. (emphasis added)

The above cited section of <u>In re Deuel</u> is concerned with <u>structural similarity</u> between chemical compounds and has nothing to do with "functional homologs" as alleged by the final Office Action. Appellants respectfully submit that, since the primers of the present

application are not structurally similar to the primers of HAUGLAND, HAUGLAND, by itself, does not establish a *prima facie* case of obviousness.

In the "Response to Arguments" (pg. 14), the final Office Action cites to M.P.E.P. § 2144.06 for allegedly supporting the proposition that the primers of Appellants' claimed methods "clearly would have represented equivalents of Haugland's primers to one of ordinary skill in the art for the purpose of practicing Haugland's method." It is prima facie obvious to combine two compositions each of which is taught by the prior art to be useful for the same purpose, in order to form a third composition to be used for the very same purpose. In re Kerkhoven, 626 F.2d 846, 850 (CCPA 1980). See also M.P.E.P. § 2144.06. However, in order to rely on equivalence as a rationale supporting an obviousness rejection, the equivalency must be recognized in the prior art, and cannot be based on applicant's disclosure or the mere fact that the components at issue are functional or mechanical equivalents. In re Ruff, 256 F.2d 590 (CCPA 1958). See also M.P.E.P. § 2144.06. Though the primers of the present application are functionally equivalent to the primers of HAUGLAND (e.g., both are used for detecting S. chartarum), there is no recognition in the prior art that Appellants' primers or probes are in any way equivalent to any of the primers or probes disclosed in any of the references cited by the final Office Action. Since it appears that the final Office Action is asserting the obviousness of Appellants' claimed primer and probes merely on their functional equivalence to the primers or probes disclosed by HAUGLAND, and not based on any

equivalence recognized in the prior art, Appellants submit that the final Office Action's equivalence argument is improper, and does not establish the obviousness of Appellants' primers and probes.

In the "Response to Arguments" section (pg. 14), the final Office Action further alleges that "the known prior art sequence disclosed in" GENBANK "renders the primers of the claimed methods obvious, because the structure of the latter is completely found within the former." Appellants traverse and submit that the mere disclosure of a genomic sequence does not offer any information as to regions that are unique for amplifying a specific target. One skilled in the art will recognize that primers from any given sequence, such as GENBANK, must be carefully designed and extensively tested to verify that the designed primers actually detect only the target organism, such as the fungus *S. chartarum*, with species specificity.

On pages 11-12, the final Office Action further cites to the disclosure of BUCK as allegedly providing evidence that Appellants' primers are obvious. Appellants submit that BUCK does not demonstrate that there is a reasonable expectation of success of achieving the claimed invention and, therefore, that the final Office Action has failed to establish a *prima facie* case of obviousness.

To make a proper rejection under 35 U.S.C. §103(a), the burden is on the Examiner to establish a *prima facie* case of obviousness. See M.P.E.P. § 2142. As one requirement for establishing a *prima facie* case of obviousness, there must be a reasonable expectation of success

of achieving the claimed invention when combining the teachings of the applied references. *In re Vaeck*, 947 F.2d 488, U.S.P.Q.2d 1438 (Fed. Cir. 1991). See M.P.E.P. § 2143. References can, thus, be modified or combined to reject a claim as *prima facie* obvious as long as there is a reasonable expectation of success. In re Merck & Co., Inc., 800 F.2d 1091, 231 USPQ 375 (Fed. Cir. 1986). See M.P.E.P. § 2143.02.

In the "Evidence Appendix" of this appeal brief, Appellants have included a copy of Dr. Linda Stetzenbach's Declaration under 37 C.F.R. § 1.132 and CV which was originally submitted on June 8, 2006. Dr. Linda Stetzenbach provides her expert opinion, based on an analysis of the HAUGLAND, BUCK and GENBANK references, that the disclosures of these references do not evidence that there was a reasonable expectation of success of deriving the primer and probe set of the present application, that includes primer (SEO ID NO: 1) 5'GTTGCTTCGGCGGGAAC3', primer (SEQ ID NO: 2) 5'TTTGCGTTTGCCACTCAGAG3', and probe (SEQ ID NO: 5) 6-FAM-5'CTGCGCCCGGATCCAGGC3'-TAMRA, for use in determining a concentration of the fungus Stachybotrys chartarum in a sample, where the primers and probe do not cross-react with other fungal species when used in combination in quantitative polymerase chain reaction. Furthermore, Dr. Linda Stetzenbach provides her expert opinion, based on an analysis of the HAUGLAND, BUCK and GENBANK references, that the disclosures of these references do not evidence that there was a reasonable expectation of success of deriving the primer and probe set of the present application, that includes primer (SEQ ID NO: 3) 5'ACCTATCGTTGCTTCGGCG3', primer (SEQ ID NO: 4)

5'GCGTTTGCCACTCAGAGAATACT3' and probe (SEQ ID NO: 5) 6-FAM5'CTGCGCCCGGATCCAGGC3'-TAM, for use in determining a concentration of the fungus

Stachybotrys chartarum in a sample, where the primers and probe do not cross-react with other

fungal species when used in combination in quantitative polymerase chain reaction.

Dr. Stetzenbach, at the time the invention was made, was the Director of the Microbiology Division of the Harry Reid Center for Environmental Studies at the University of Nevada, Las Vegas and, thus, was the inventors' supervisor. Dr. Stetzenbach was also the Project Director for the U.S. Department of Energy award that funded the study from which the invention resulted. The National Science Foundation additionally provided funds to complete the project. The invention was also part of Appellant Dr. Patricia Cruz-Perez's dissertation, for which Dr. Stetzenbach was Dr. Cruz-Perez's advisor. Dr. Stetzenbach, however, is no longer Director of the Microbiology Division of the Harry Reid Center for Environmental Studies, but is now a Professor in the Environmental and Occupational Health Program of the School of Public Health of the University of Nevada, Las Vegas. Dr. Stetzenbach does not have any financial stake in this invention, or in the issuance of this patent application as a patent. Dr. Stetzenbach's CV is attached as an exhibit to the Rule 132 declaration as evidence of her qualifications and expertise in environmental microbiology and, particularly, in the use of PCR for the detection of environmental microorganisms such as *S. chartarum*.

In view of the facts, opinion and evidence contained in the attached declaration by Dr. Stetzenbach, Appellants respectfully submit that the HAUGLAND, BUCK and GENBANK

references do not show that there was a reasonable expectation of success of producing the claimed invention. Since the cited references do not show that there was a reasonable expectation of success, the final Office Action has failed to establish a *prima facie* case of obviousness.

In the "Response to Arguments" section (pg. 15), the final Office Action asserts that Dr. Stetzenbach's testimony, contained in the Declaration under 37 C.F.R. § 1.132, stating that "extensive research would have to be conducted to find primers that will not cross-react with other organisms," is "not persuasive." The final Office Action further asserts the following:

Homology searches in sequence databases was routinely used in the art at the time the invention of the instant application was made (for example, the commonly used BLAST algorithm was introduced in 1990). This analysis could have been performed rather easily over the internet. For example, the sequence alignments depicted in the rejection above took less than 30 minutes for the examiner to produce using the BLAST algorithm on the National Center for Biotechnology Information (NCBI) web site...this would not have been considered 'extensive' research by one of ordinary skill in the art in 2001.

Appellants respectfully submit there is no merit to the final Office Action's allegations, because finding primers that will not cross-react with other (fungal) species requires extensive research that not only includes sequence homology comparisons using a method such as the BLAST algorithm, but also the following:

1) sequence comparison results are analyzed visually to locate areas of homology and heterogeneity in order to find unique region(s) in the target sequence;

- 2) the unique region(s) are imported into a primer design software program for the design of potential primer and probe sequences, and primer design criteria are modified to improve the chances of obtaining suitable primer sets;
 - 3) numerous potential primer and probe sets are selected and (commercially) synthesized;
- 4) several strains of the target organism as well as related and distant organisms are identified, ordered commercially, and grown in the laboratory;
 - 5) DNA is extracted and purified from all target and non-target microorganisms;
- 6) the concentration of the extracted DNA is measured using ethidium bromide staining and ultraviolet light (UV) visualization (or similar method);
 - 7) the designed primer/probe set(s) are tested experimentally with the target organism;
 - 8) the amplification assay is optimized and primer set(s) are accepted or rejected; and
- 9) primer validation is finalized by performing PCR assays with the DNA extracted from related and distant organisms. For example, Suzuki *et al.* (page 4607, 2nd sentence, (ii) Specificity section) examined "possible cross-reactivity of the primers" by looking at "the formation of PCR products after 25 cycles in three-step PCRs." (Suzuki, M.T., L.T. Taylor, and E.F. DeLong. 2000. Quantitative analysis of small-subunit rRNA genes in mixed microbial populations via 5'-nuclease assays. Applied and Environmental Microbiology. 66:4605-4614). In the work of Greisen *et al.* (page 335, 5th sentence of *Abstract*), primers were tested with "DNA from...124 different species of bacteria..." (Greisen, K., M. Loeffelholz, A. Purohit, and D. Leong. 1994. PCR primers and probes for the 16S rRNA

gene of most species of pathogenic bacteria, including bacteria found in cerebrospinal fluid.

Journal of Clinical Microbiology. 32:335-351). Appellants respectfully submit that the above-noted research process for finding primers that will detect *S. chartarum*, but will also not cross-react with other fungal species, would clearly have represented "extensive" research in 2001.

In the "Response to Arguments" section (pg. 16), the final Office Action asserts that Dr. Stetzenbach's testimony on pg. 4 of the Declaration under 37 C.F.R. § 1.132 is "not persuasive." This section of Dr. Stetzenbach's testimony submits that, "while the criteria for primer selection for sequencing reactions are flexible as demonstrated by BUCK, selection of optimal primers for PCR follows very stringent criteria and is the factor that is least predictable to troubleshoot." The final Office Action asserts that "[t]his is not persuasive because in both PCR and sequencing reactions, the function of the primer is to hybridize...as discussed in the preceding paragraph, there were readily available tools prior to 2001 for comparing sequences to find regions unique for a target of interest, and testing primers and probes for specificity would not have involved 'extensive' research."

Appellants submit that this statement of the final Office Action does not have any merit because numerous replicates of all of the amplifications involved in primer validation must be performed to determine the variability of the data and to scientifically verify that tested primers and probes can identify the fungus *Stachybotrys chartarum* in a sample without cross-reacting with other fungal species. It takes approximately 6 months for proper design and

validation of primers and probes for a target organism such as the one performed by the Appellants. In addition, the design and evaluation of PCR primers and probes is the sole subject of numerous peer-reviewed research papers that require an adequate, if not extensive, amount of experimentation for publication. For example, Nadkarni et al. (Page 257, 2nd sentence of Abstract) determined that "Broad specificity of the universal detection system was confirmed by testing DNA isolated from 34 bacterial species encompassing most of the groups of bacteria outlined in Bergey's Manual of Determinative Bacteriology." (Nadkarni, M.A., F.E. Martin, N.A. Jacques, and N. Hunter. 2002. Determination of bacterial load by realtime PCR using a broad-range (universal) probe and primers set. Applied and Environmental Microbiology. 148:257-266). Rintala and Nevalainen recently published a study with the sole aim of (page 745, last sentence of the *Introduction*) developing "a quantitative PCR assay for detection of streptomycetes in environmental samples" (Rintala, H. and A. Nevalainen. 2006. Quantitative measurement of streptomycetes using real-time PCR. Journal of Environmental Monitoring. 8:745-749). On page 746, 3rd sentence of the Design and testing of Streptomyces-specific primers section, Rintala indicates that: "Several primer pairs and TagMan probes were designed. The specificity of the primers and probes was first validated through BLAST searching against the GenBank database (http://www.ncbi.nlm.nih.gov/blast) and after that the primers were experimentally tested with conventional PCR for amplification of Streptomyces and other species." Furthermore, on page 746, 1st sentence of the Real-time PCR assay for streptomycetes section, Rintala specifies: "Based on the results of the testing

with conventional PCR, one primer pair was chosen and the specificity of the primer set and the corresponding TaqMan probe was tested with real-time PCR assay."

In view of the remarks above, Appellants submit that the final Office Action has not established a *prima facie* case of obviousness. Reversal of the rejection of claims 19, 20 and 22-27 is, therefore, respectfully requested.

VIII. CONCLUSION

For at least the foregoing reasons, it is respectfully requested that the Examiner's rejections of claims 18 and 21 under 35 U.S.C. §102(b) and claims 19, 20 and 22-27 under 35 U.S.C. §103 be REVERSED.

Respectfully submitted,

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Date: February 14, 2007

CLAIMS APPENDIX

THE APPEALED CLAIMS

The claims on appeal are as follows:

18. A method for identifying and quantifying the presence of the fungus *Stachybotrys* chartarum in a collected sample, comprising:

obtaining a primer set and probe that is specific for the fungal species *Stachybotrys* chartarum;

collecting the sample from the environment;

extracting the sample's DNA;

obtaining DNA standards from a culture of Stachybotrys chartarum;

determining the concentration of Stachybotrys chartarum spores in the DNA standards;

amplifying by polymerase chain reaction each of the DNA standards and the collected sample's DNA using the obtained primer set and probe; and

comparing amplification plots obtained by polymerase chain reaction of each of the DNA standards and the collected sample's DNA to obtain an indication of the presence of the fungus *Stachybotrys chartarum* in the collected sample and a concentration of the fungus *Stachybotrys chartarum* in the collected sample.

- 19. The method of claim 18, wherein the primer set and probe comprises:
 - a forward primer comprising a base sequence (SEQ ID NO: 1)

5'GTTGCTTCGGCGGGAAC3';

a reverse primer comprising a base sequence (SEQ ID NO: 2)

5'TTTGCGTTTGCCACTCAGAG3'; and

a probe comprising a base sequence (SEQ ID NO: 5) 6-FAM-

5'CTGCGCCCGGATCCAGGC3'-TAMRA, wherein the forward primer, reverse primer and probe do not cross-react with other fungal species when used in combination in polymerase chain reaction.

- 20. The method of claim 18, wherein the primer set and probe comprises:
 - a forward primer comprising a base sequence (SEQ ID NO: 3)

5'ACCTATCGTTGCTTCGGCG3';

a reverse primer comprising a base sequence (SEQ ID NO: 4)

5'GCGTTTGCCACTCAGAGAATACT3'; and

a probe comprising a base sequence (SEQ ID NO 5) 6-FAM-

5'CTGCGCCCGGATCCAGGC3'-TAMRA, wherein the forward primer, reverse primer and probe do not cross-react with other fungal species when used in combination in polymerase chain reaction.

- 21. The method of claim 18, wherein the concentration of *Stachybotrys chartarum* spores in the DNA standards is determined via direct total count of the *Stachybotrys chartarum* spores in the DNA standards.
- 22. A method for identifying and quantifying the presence of the fungus *Stachybotrys* chartarum in a collected sample, comprising:

obtaining a primer set and probe that is specific for the fungal species *Stachybotrys* chartarum,

wherein the obtained primer set and probe comprises:

a forward primer comprising a base sequence (SEQ ID NO: 1)

5'GTTGCTTCGGCGGGAAC3';

a reverse primer comprising a base sequence (SEQ ID NO: 2)

5'TTTGCGTTTGCCACTCAGAG3'; and

a probe comprising a base sequence (SEQ ID NO: 5) 6-FAM-

5'CTGCGCCCGGATCCAGGC3'-TAMRA; and

employing quantitative polymerase chain reaction, using the obtained primer and probe set, to determine a concentration of the fungus *Stachybotrys chartarum* in the collected sample, wherein the forward primer, reverse primer and probe do not cross-react with other fungal species when used in combination in the quantitative polymerase chain reaction.

23. The method of claim 22, wherein employing quantitative polymerase chain reaction further comprises:

extracting the collected sample's DNA;

obtaining one or more DNA standards from a culture of Stachybotrys chartarum;

determining a concentration of *Stachybotrys chartarum* spores in each of the one or more DNA standards;

amplifying by polymerase chain reaction each of the one or more DNA standards and the collected sample's DNA using the obtained primer set and probe; and

comparing amplification plots obtained by polymerase chain reaction of each of the one or more DNA standards and the collected sample's DNA to determine the concentration of the fungus *Stachybotrys chartarum* in the collected sample.

- 24. The method of claim 23, wherein the concentration of *Stachybotrys chartarum* spores in each of the one or more DNA standards is determined via direct total count of the *Stachybotrys chartarum* spores in the DNA standards.
- 25. A method for identifying and quantifying the presence of the fungus *Stachybotrys* chartarum in a collected sample, comprising:

obtaining a primer set and probe that is specific for the fungal species *Stachybotrys* chartarum,

wherein the obtained primer set and probe comprises:

a forward primer comprising a base sequence (SEQ ID NO: 3)

5'ACCTATCGTTGCTTCGGCG3';

a reverse primer comprising a base sequence (SEQ ID NO: 4)

5'GCGTTTGCCACTCAGAGAATACT3'; and

a probe comprising a base sequence (SEQ ID NO: 5) 6-FAM-

5'CTGCGCCCGGATCCAGGC3'-TAM; and

employing quantitative polymerase chain reaction, using the obtained primer and probe set, to determine a concentration of the fungus *Stachybotrys chartarum* in the collected sample, wherein the forward primer, reverse primer and probe do not cross-react with other fungal species when used in combination in the quantitative polymerase chain reaction.

26. The method of claim 25, wherein employing quantitative polymerase chain reaction further comprises:

extracting the collected sample's DNA;

obtaining one or more DNA standards from a culture of Stachybotrys chartarum;

determining a concentration of Stachybotrys chartarum spores in each of the one or more

DNA standards;

amplifying by polymerase chain reaction each of the one or more DNA standards and the collected sample's DNA using the obtained primer set and probe; and

comparing amplification plots obtained by polymerase chain reaction of each of the one or more DNA standards and the collected sample's DNA to determine the concentration of the fungus *Stachybotrys chartarum* in the collected sample.

27. The method of claim 26, wherein the concentration of *Stachybotrys chartarum* spores in each of the one or more DNA standards is determined via direct total count of the *Stachybotrys chartarum* spores in the DNA standards.

EVIDENCE APPENDIX

Dr. Linda Stetzenbach's Declaration under 37 C.F.R. § 1.132 and CV. This Declaration was originally submitted with Appellants' Amendment filed June 8, 2006.



Patent Attorney's Docket No. <u>0001-00001CON1</u>

Certificate of Mailing

I hereby certify that this correspondence is being addressed as set out in 37 CFR §1.1(a) and deposited with the U.S. Postal Service with sufficient postage as first class mail on June 8, 2006.

Typed or printed name of person signing this certificate: <u>Tony M. Cole</u>

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE			
In re Patent Application of)		
Patricia CRUZ-PEREZ et al.) Group Art Unit: 1637		
Application No.: 10/804,339) Examiner: Samuel C. Woolwine		
Filed: March 19, 2004)		
For: METHOD FOR DETECTION OF Stachybotrys chartarum IN PURE CULTURE AND FIELD SAMPLES USING QUANTITATIVE POLYMERASE CHAIN REACTION))))		
	,		

Signature _

DECLARATION UNDER 37 C.F.R. § 1.132

Mail Stop: Amendments Commissioner for Patents P.O. Box 1450 Alexandria, Virginia 22313-1450

Sir:

I, Dr. Linda Stetzenbach, have reviewed the Office Action dated February 8, 2006, and the references cited by the Examiner therein for rejecting the claims of the present application, and hereby declare that:

- 1. I received my Ph.D. from the University of Arizona in Microbiology in 1986. I have worked in the field of environmental microbiology for 19 years, and have authored numerous papers, which are detailed on my attached CV. These include several peer-reviewed publications and book chapters on the use of polymerase chain reaction (PCR) for the detection of microorganisms of indoor air quality concern. Based on my education and years of experience, I have specific expertise in environmental microbiology, and particularly, in the use of PCR for the detection of environmental microorganisms, such as, for example, the fungal species *Stachybotrys chartarum*.
- 2. I have analyzed the reference "Quantitative Measurement of Stachybotrys chartarum conidia Using Real Time Detection of PCR Products with the TaqManTM Fluorogenic Probe System," R.A. Haugland et al., Molecular and Cellular Probes (1999) 13: 329-340 (hereinafter "HAUGLAND") cited by the Office Action and state the following:
 - a. HAUGLAND discloses a method for detecting the fungal species S. chartarum that includes a primer and probe set that is species specific. However, the primers and probe disclosed by HAUGLAND do not include the primer and probe sets recited in Applicants' claims. HAUGLAND, thus, does not disclose or suggest the primer and probe sets disclosed and claimed in the present application.
- 3. I have analyzed the genomic sequences disclosed in GenBank GI: 3420911 (hereinafter "GENBANK"). Applicants' SEQ ID NOs. 1-5 are homologous to portions of the *Stachybotrys chartarum* genomic sequence deposited in GenBank with accession number AF081469. Several researchers have sequenced portions of the *Stachybotrys chartarum* genome and many of these have been published and/or submitted to DNA sequence repositories such as GenBank. The mere publication of a portion or the complete genome of an organism, however, does not make it a suitable region for PCR amplification. Species-specific primers and probes can theoretically be derived from any

DNA sequence; however that unique region in the target genome needs to be located first. The genomic sequence of the organism of interest would have to be compared with all the sequence information available in the genetic repositories and scientific literature databases in order to locate a <u>unique</u> region in the target genome. <u>Extensive</u> research and validation would have to then be conducted by means of specificity testing in order to design PCR primers and probes that are ultimately specific for the target organism and will not cross-react with other organisms.

- 2. I have analyzed the reference "Design Strategies and Performance of Custom DNA Sequencing Primers," Bio Techniques 27:528-536 (September 1999) (hereinafter "BUCK") cited by the Office action and submit the following:
 - BUCK discloses a methodology for designing non-species specific a. primers for the purpose of deciphering the sequence of all or part of a genome. The purpose of the sequencing reaction disclosed in BUCK is to decipher the sequence of all or part of a genome. In enzymatic sequencing reactions, priming of DNA synthesis is achieved by the use of a synthetic oligonucleotide (primer) complementary to a specific sequence on the DNA template strand (Sambrook, J., E.F. Fritsch, and T. Maniatis. 1989. DNA Sequencing, pp. 13.1-13.104. In Molecular Cloning. A Laboratory Manual. 2nd Edition. Cold Spring Harbor Laboratory Press, New York). BUCK demonstrates that nearly any primer sequence that is complementary to the template DNA is suitable for a sequencing reaction. BUCK's primers are sequencing primers which are by definition nonspecific primers designed for the purpose of deciphering the sequence of all or part of a genome. These primers are normally designed with very little stringency so that they can bind almost any portion of DNA, even segments that have very little homology to the primer sequence, thus producing short strands of DNA of the genome of interest that may be pieced together for the purpose of sequencing part or all of the genome. Therefore, these sequencing primers of BUCK cannot be

utilized for the accurate and specific detection and amplification of target organisms.

- b. For PCR detection of a target microorganism, knowledge of a unique DNA sequence is required, and then primers are selected that are complementary to DNA regions flanking the target sequence, and then tested to ensure that they do not cross-react with non-target DNA. Extensive research and validation would have to then be conducted by means of species specificity testing in order to design PCR primers and probes that are ultimately specific for the target organism and will not cross-react with other organisms. While the criteria for primer selection for sequencing reactions are flexible as demonstrated by BUCK, selection of optimal primers for PCR follows very stringent criteria and is the factor that is least predictable to troubleshoot (Ausubel, F.M., R. Brent, R.E. Kingston, D.D. Moore, J.G. Seidman, J.A. Smith, and K. Struhl. 1995. The Polymerase Chain Reaction, pp. 15.0.1-15.7.11. In Current Protocols in Molecular Biology. John Wiley & Sons, Inc., New York). Therefore, sequencing primers, such as those disclosed by BUCK, would not be utilized by those skilled in the art for the purpose of designing PCR primers and probes for the accurate and specific detection and amplification of target organisms. BUCK, thus, has no teaching that is relevant to the selection of a primer from a sequence of bases for use in identifying a specific target organism, such as S. chartarum.
- 3. In view of the above, my conclusion after analyzing the HAUGLAND, BUCK and GENBANK references is that the disclosures of these references, taken alone, or in combination, do not evidence that there was a reasonable expectation of success of deriving the primer and probe sets of the present application that includes primer (SEQ ID NO: 1) 5'GTTGCTTCGGCGGGGAAC3', primer (SEQ ID NO: 2) 5'TTTGCGTTTGCCACTCAGAG3', and probe (SEQ ID NO: 5) 6-FAM-5'CTGCGCCCGGATCCAGGC3'-TAMRA; or primer (SEQ ID NO: 3)

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(And

5'ACCTATCGTTGCTTCGGCG3', primer (SEQ ID NO: 4)
5'GCGTTTGCCACTCAGAGAATACT3' and probe (SEQ ID NO: 5) 6-FAM5'CTGCGCCCGGATCCAGGC3'-TAM; for use in determining the concentration of the fungus *Stachybotrys chartarum* in a sample, wherein the primers and probe <u>do not cross-react</u> with other fungal species when used in combination in quantitative polymerase chain reaction.

4. I further declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and, further, that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issuing therefrom.

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Date:	June 7, 2006	Signature:	

LIST OF EXHIBITS

A: CV of Dr. Linda Stetzenbach

EXHIBIT A

LINDA D. STETZENBACH

Professor, Environmental and Occupational Health Program
School of Public Health
August 1, 2005-present
University of Nevada, Las Vegas
4505 South Maryland Parkway, Box 454009
Las Vegas, NV 89154-4009
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Previous Employment:

Director - Microbiology Division, August 1, 1987 - July 31, 2005, Harry Reid Center for Environmental Studies, University of Nevada, Las Vegas

Education:

B.S. - Microbiology, January 1970 Department of Microbiology and Immunology University of Arizona, Tucson, AZ 85721

M.S. - Microbiology, May 1984 Department of Microbiology and Immunology University of Arizona, Tucson, AZ 85721 **Ph.D.** - Microbiology, May 1986 Department of Microbiology and Immunology University of Arizona, Tucson, AZ 85721

Post-doctoral fellow, April 1986 - July 1987 Department of Veterinary Science University of Arizona, Tucson, AZ 85721

Approximately ten years of clinical/medical microbiology experience (1970-1981) including 6 years at the Veterans' Administration Medical Center, Tucson, AZ (1975-1891).

Publications

Alvarez, A.J., M.P. Buttner, and L.D. Stetzenbach. 1995. PCR for Bioaerosol Monitoring: sensitivity and environmental interference. Appl. Environ. Microbiol. 61:3639-3644.

Alvarez, A.J., M.P. Buttner, G.A. Toranzos, E.A. Dvorsky, A. Toro, T.B. Heikes, L.E. Mertikas, and L.D. Stetzenbach. 1994. The Use of Solid-phase Polymerase Chain Reaction for the Enhanced Detection of Airborne Microorganisms. Appl. Environ. Microbiol. 60:374-376.

Buttner, M.P., P. Cruz, **L.D. Stetzenbach**, A.K. Klima-Comba, V.L. Stevens, and P.A. Emanuel, 2004. Evaluation of the Biological Sampling Kit for Large Area Microbial Surface Sampling. Appl. Environ. Microbiol. 70:7040-7045.

Buttner, M.P., P. Cruz, L.D. Stetzenbach, A.K. Klima-Comba, V.L. Stevens, and T.D. Cronin. 2004. Determination of the Efficacy of Two Building Decontamination Strategies by Surface Sampling with Culture and Quantitative PCR Analysis. Appl. Environ. Microbiol. 70:4740-4747.

Buttner, M.P., P. Cruz-Perez, L.D. Stetzenbach, P.J. Garrett, and A.L. Luedtke. 2002. Measurement of Airborne Fungal Spore Dispersal from Three Types of Flooring Materials. Aerobiologia, 18:1-11.

Buttner, M.P., P. Cruz-Perez, and L.D. Stetzenbach. 2001. Enhanced Detection of Surface-associated Bacteria in Indoor Environments using Quantitative PCR. Appl. Environ. Microbiol.67:2564-2570.

Buttner, M.P., P.Cruz-Perez, P.J. Garrett, and L.D. Stetzenbach. 1999. Dispersal of Fungal Spores from Three types of Air Handling System Duct Material. Aerobiologia. 15:1-8.

Buttner, M.P., A.J. Alvarez, L.D. Stetzenbach, and G.A. Toranzos, 1997. PCR Detection of Airborne Microorganisms, pp. 145-158. *In G.A. Toranzos* (ed.), Environmental Applications of Nucleic Acid Amplification Techniques, Technomic Publishing Co., Lancaster.

Buttner, M.P. and L.D. Stetzenbach. 1996. The Use of an Experimental Room for Monitoring of Airborne Concentrations of Microorganisms, Glass Fibers, and Total Particles, p. 75-86. *In* B.A. Tichenor (ed.), Characterizing Sources of Indoor Air Pollution and Related Sink Effects. ASTM 1287, American Society for Testing and Materials, West Conshohocken, PA.

Buttner, M.P. and **L.D. Stetzenbach**. 1993. Monitoring of Fungal Spores in an Experimental Indoor Environment to Evaluate Sampling Methods and the Effects of Human Activity on Air Sampling. Appl. Environ. Microbiol. **59:**219-226.

Buttner, M.P. and **L.D. Stetzenbach**. 1991. Evaluation of Four Aerobiological Sampling Methods for the Retrieval of Aerosolized *Pseudomonas syringae*. Appl. Environ. Microbiol. **57:**1268-1270.

Craner, J., and L.D. Stetzenbach. 1999. Diagnosing the Cause of a >Sick Building=: a case study of an epidemiological and microbiological investigation. *In* E. Johanning (ed.), Bioaerosols, Fungi and Mycotoxins: Health Effects, Assessment, Prevention, and Control. Eastern New York Occupational and Environmental Health Center, Albany, NY.

Cross-Smiecinski, A. and L.D. Stetzenbach. 1994. Quality Planning for the Life Science Researcher: Meeting Quality Assurance Requirements. CRC Press Inc., Boca Raton, FL.

Cruz, P. and L.D. Stetzenbach. 2004. Specific Detection of Fungi Associated with SBS when using Quantitative Polymerase Chain Reaction. Adv. App. Microbiol. 55:437-449.

Cruz, P., J.L. Henry, A.K. Klima-Comba, and **L.D. Stetzenbach**. 2002. Air and Surface Sampling Measurements of Fungal Contaminants in Indoor Environments. Proceedings of Indoor Air 2002, p. 420-424.

Cruz-Perez, P., M.P. Buttner, and **L.D. Stetzenbach**. 2001. Specific Detection and Quantitation of *Aspergillus fumigatus* in Pure Culture using Quantitative Polymerase Chain Reaction. Mol. Cellular Probes. 15:81-88.

Cruz-Perez, P., M.P. Buttner, and L.D. Stetzenbach. 2001. Specific Detection of *Stachybotrys chartarum* in Pure Culture using Quantitative Polymerase Chain Reaction. Mol. Cellular Probes. 15: 129-138.

Lawson, C.N., H.S. Bailey, C.A. Callaway, L. Harriman III, R.H. Lewis, C.M. McDonald, L.D. Stetzenbach, W. Thomann. 2005. Minimizing Indoor Mold Problems through Management of Moisture in Building systems. American Society of Heating, Refrigerating and Air-conditioning Engineers, Inc., Atlanta, GA.

Lighthart, B. and L.D. Stetzenbach. 1994. Distribution of Microbial Bioaerosol, p. 68-98. *In* B. Lighthart and A.J. Mohr (ed.), Atmospheric Microbial Aerosols, Theory and Applications, Chapman and Hall, New York, NY.

Maki, L.R., S-H Shen, R.C. Bergstrom, and L.D. Stetzenbach. 1985. Diagnosis of *Corynebacterium pseudotuberculosis* in Sheep, Using an Enzyme-linked Immunosorbent Assay. Am. J. Vet. Res. 46: 212-214.

Metha, S.K., D.M. Bell-Robinson, T.O. Groves, **L.D. Stetzenbach**, and D. L. Pierson. 2000. Evaluation of Portable Air Samplers for Monitoring Airborne Culturable Bacteria. Am. Indust. Hyg. Assoc. J. **61**: 850-854.

Sterling, C.R., R.M. Kutob, M.J. Gizinski, M. Verastegui, and **L.D. Stetzenbach**. 1988. *Giardia* Detection Using Monoclonal Antibodies Recognizing Determinants of *In Vitro* Derived Cyst, p. 219-222. *In* P.M. Wallis and B.R. Hammond (ed.), Advances in *Giardia* Research. University of Calgary Press, Calgary.

Stetzenbach, L.D. and M.P. Buttner. 2005. Sampling and Analysis for Mold Contamination in the Indoor Environment. *In J.Herz*, and K. Taylor (ed.), Toxic Mold Litigation (on-line version: www.lawyersandjudges.com), Lawyers and Judges Publishing Co., Tucson, AZ.

Stetzenbach, L.D. 2005. Bacteriology of Air, p. 185-194. *In* S.P. Borriello, P.R. Murray, and G. Funke (ed.), Topley and Wilson's Microbiology and Microbial Infections, 10th Edition, Bacteriology, Volume 1, Health Sciences Publishing, London.

Stetzenbach, L.D., H.A. Amman. E. Johanning, G. King, R.J. Shaughnessy. 2004. Microorganisms, Mold, and Indoor Air Quality. American Society for Microbiology, Washington, DC.

Stetzenbach, L.D., M.P. Buttner, and P. Cruz. 2004 Detection and Enumeration of Airborne Biocontaminants. Cur. Op. Biotechnol. 15:170-174.

Stetzenbach, L.D., and M.V. Yates. 2003. The Dictionary of Environmental Microbiology. Academic Press, San Diego, CA.

Stetzenbach, L.D. 2002. Introduction to Aerobiology, p. 801-813. *In* C. J. Hurst, R.L. Crawford, G. Knudsen, M. McInerney, and L.D. Stetzenbach, (ed.), Manual of Environmental Microbiology, second edition. ASM Press, Washington, DC.

Stetzenbach, L.D. 2002. Enhanced Detection of Airborne Microbial Contaminants, p. 1130-1136. *In* Gabriel Bitton (ed.), Encyclopedia of Environmental Microbiology. John Wiley and Sons, Inc., New York, NY.

Stetzenbach, L.D., and M.P. Buttner. 2000. Airborne Microorganisms and Indoor Air Quality, pp. 116-125. *In J.* Lederberg (ed.), Encyclopedia of Microbiology, second edition. Academic Press, San Diego, CA.

Stetzenbach, L.D. 1998. Microorganisms and Indoor Air Quality. Clin. Microbiol. Newsletter. **20:**157-160.

Stetzenbach, L.D. 1997. Introduction to Aerobiology, p. 619-628. *In* C.J. Hurst, G. Knudsen, M. McInerney, M.V. Walter, and L.D. Stetzenbach, (ed.), Manual of Environmental Microbiology, ASM Press, Washington, DC.

Stetzenbach, L.D., A.J. Alvarez, and M.P. Buttner. 1996. The Use of Polymerase Chain Reaction (PCR) to Enhance Bioaerosol Monitoring. *In* D.A. Berg (ed.), Proceedings of the 1994 ERDEC Scientific Conference on Chemical and Biological Defense Research. Aberdeen Proving Ground, MD.

Stetzenbach, L.D. 1992. Airborne Microorganisms, p. 53-66. *In J. Lederberg (ed.)*, Encyclopedia of Microbiology. Academic Press, Harcourt Brace Joanovich Publishers, San Diego. CA.

Stetzenbach, L.D., M.P. Buttner, and J.R. Meldrum. 1992. Indoor Air Investigations and Controlled Laboratory Studies using a Variety of Aerobiological Samplers. Institute of Environmental Sciences, Mount Prospect, IL.

Stetzenbach, L.D., B. Lighthart, R.J. Seidler, and S.C. Hern. 1992. Factors Influencing the Dispersal and Survival of Aerosolized Microorganisms, p. 455-465. *In M. Levin*, R.J. Seidler, M.

Rogul, and H.A.P. Pritchard (ed.), Microbial Ecology: Principles, Methods, and Applications. McGraw-Hill, Inc., New York, NY.

Stetzenbach, L.D., S.C. Hern, and R.J. Seidler. 1992. Field Sampling Design and Experimental Methods for the Detection of Airborne Microorganisms, p. 543-555. *In* M. Levin, R.J. Seidler, M. Rogul, and H.A.P. Pritchard (ed.), Microbial Ecology: Principles, Methods, and Applications. McGraw-Hill, Inc., New York, NY.

Stetzenbach, L.D., L. M. Kelley, and N.A. Sinclair. 1986. Isolation, Identification, and Growth of Well-water Bacteria. Groundwater. 24: 6-10.

Yates, M.V. L.D. Stetzenbach, C.P. Gerba, and N.A. Sinclair. 1990. The Effect of Indigenous Bacteria on Virus Survival in Ground Water. J. Environ. Sci. Health. A25: 81-100.

National and International Meetings (2005-1994)

2005:

Invited Presentations

Stetzenbach, L.D., M. Buttner, and P. Cruz. Surface Sampling for Biocontaminants in Indoor Environments. <u>Biological Background Forum</u>. Department of Homeland Security and Los Alamos National Laboratory, July 27, Santa Fe, NM.

Mold in Indoor Environments – Background Information and Monitoring Practices. <u>Mold the Building Environment</u>. American Society for Heating, Refrigeration, and Air Conditioning Engineers, ASHRAE Webcast April 13, Washington DC.

Bioaerosol and Surface Monitoring for Biothreat Agents. <u>Assessing Anthrax Detection Methods</u>, Government Reform Committee Subcommittee on National Security, Emerging Threats and International Relations, April 5, Washington, DC.

Buttner, P. Cruz, **L.D. Stetzenbach**, A.K. Klima-Comba, V.L. Stevens, and P.A. Emanuel. Evaluation of the Biological Sampling Kit (BiSKit). <u>Advanced Sample Collection Methods and Technologies</u>, National Conference on Environmental Sampling for Bio-threat Agents. January 28, Baltimore, MD.

Cruz, P. M.P. Buttner, and **L.D. Stetzenbach**. Surface Biocontaminant Detection Strategies. <u>Surface Environmental Sample Collection</u>, National Conference on Environmental Sampling for Bio-threat Agents. January 27, Baltimore, MD.

Presentations

Stetzenbach, L.D. Indoor Environmental Quality in Office Buildings and Educational Facilities. Nevada Public Health Association Conference, October 13-14, Lake Tahoe, NV

Cruz, P., L.D. Stetzenbach, A.K. Klima-Comba, V.L. Stevens, V.A. Castro, C.M. Ott, D.L. pierson. Enhanced Detection of Fungi in the Spacecraft Environment. Abstr. Y-051. The 105th General Meeting of the American Society for Microbiology. June 7, Atlanta, GA.

King, G.M., L.D. Stetzenbach, A.K. Klima-Comba. Analysis of Cultivable Airborne Bacteria from an Altitude Gradient on Kilauea and Mauna Loa Volcanoes (Hawai'i). Abstr. N-094. The 105th General Meeting of the American Society for Microbiology. June 6, Atlanta, GA.

Klima-Comba, A.K., J.L. Henry, **L.D. Stetzenbach**, G.M. King. Sampling Methodologies for Monitoring Outdoor Culturable Airborne Fungi from Five Locations on Hawai'i. Abstr. Q-003. The 105th General Meeting of the American Society for Microbiology. June 6, Atlanta, GA.

Ott, C.M., V.A. Castro, V.J. Bassinger, S.L. Fontenot, R.J. Bruce, P. Cruz, L.D. Stetzenbach, D.L. Pierson. A Comprehensive Characterization of Microorganisms and Allergens in Spacecraft Environment. NASA Bioastronautics Investigators' Workshop, January 10-12, Galveston, TX.

Session Organizer:

<u>Surface Environmental Sample Collection</u>, National Conference on Environmental Sampling for Bio-threat Agents. January 27, Baltimore, MD.

2004:

Invited Presentations

Methods for Monitoring Mold in Indoor Environments. <u>Mold-related Health Effects: Clinical, Remediation Worker Protection, and Biomedical Research Issues</u>. Society for Occupational and Environmental Health, and the National Institute for Environmental Health Science. June 28-29, Washington, DC.

Monitoring of Biocontaminants in Indoor Environments. <u>First International Conference on Fate of Biological Agents</u>, June 9, Williamsburg, VA.

Mold Contamination in Indoor Environments. National Business Institute, April 22, Las Vegas, NV.

Sampling Methods on Various Surfaces using Simulant Organisms. Workshop on Standards and Policies for Decontaminating Public Facilities Affected by Exposure to Harmful Biological Agents, National Academy of Science, January 29, Washington, DC.

Natural Background. Workshop on Standards and Policies for Decontaminating Public Facilities Affected by Exposure to Harmful Biological Agents, National Academy of Science, January 29, Washington, DC.

Mold Contamination in Indoor Environments. Lorman Educational Seminar Series, January 22, Las Vegas, NV.

Presentations

Burns-Savage, N., P.J. Garrett, **L.D. Stetzenbach**, and C. Vanier. Functional Operation of the Collison Nebulizer in Controlled Bioaerosol Experiments utilizing an Environmental Chamber, Abstr. 2602. The 104th General Meeting of the American Society for Microbiology. May 25. New Orleans, LA.

Miller, R.L., J.L. Henry P.J. Garrett and **L.D. Stetzenbach**. Abstr. Q130. Rapid Detection of *Stachybotrys chartarum*, *Aspergillus niger* and Trichothecenes in Laboratory Isolates and Environmental Samples. 104th General Meeting of the American Society for Microbiology. May 25, New Orleans, LA.

2003:

Invited Presentations

Bioaerosols: have recent events changed our perspective on breathing free? Southern California Branch of the American Society for Microbiology, November 8, San Diego, CA.

Overview of Mold Assessment II. 5th International Conference on Bioaerosols, Fungi, Bacteria, Mycotoxins, and Human Health, September 11, Saratoga Springs, NY.

Mold Colonization and Transport in Duct Systems. <u>Impacts of Duct Systems on Indoor Air Quality</u>, American Society of Heating, Refrigeration and Air Conditioning Engineers (ASHRAE) Annual Meeting, June 29, Kansas City, MO.

Fungal Contamination in Indoor Environments. National Environmental Health Association, 67th Annual Educational Conference, June 9, Reno, NV.

Mold Contamination in Indoor Environments. <u>The Mold Challenge in Nevada</u>, National Business Institute Symposium, June 3, Las Vegas, NV.

Mold Contamination in Indoor Environments. DriEaz Restorative Drying Symposium, May 3, Las Vegas, NV.

Hot Issues - sampling strategy and data interpretation. Mealey=s Mold Litigation Conference 2003, February 3, San Diego, CA.

Presentations

Buttner, M.P., P, Cruz, A.K. Klima-Comba, L.D. Stetzenbach, and P. Emanuel. Evaluation of a New Surface Sampling Method for the Detection of Biocontaminants. Abstr. Q-220, Proceedings of the 103rd General Meeting of the American Society for Microbiology, May 20, Washington, DC.

Fungal Biocontamination and Enhanced Detection using Quantitative Polymerase Chain Reaction. <u>Fungal Biocontaminants in Indoor Environments</u>, American Society for Microbiology Colloquium. 103rd General Meeting of the American Society for Microbiology, May 20, Washington DC.

Buttner, M.P., P. Cruz, A. K. Klima-Comba, **L.D. Stetzenbach**, and P. Emanuel. Validation of the BiSKit Sampler for the Detection of Microorganisms on Surfaces. <u>Biodefense Research Conference</u>, American Society for Microbiology, March 10, Baltimore, MD.

Cruz, P., M.P. Buttner, A.K. Klima-Comba, **L.D. Stetzenbach**, and T.D. Cronin. Utilization of a Room-Sized Experimental Chamber for Efficacy Testing of a Decontamination Product. . <u>Biodefense Research Conference</u>, Abstr. 322, American Society for Microbiology, March 10, Baltimore, MD.

Buttner, M.P., P. Cruz, A.K. Klima-Comba, **L.D. Stetzenbach**, and P. Emanuel. Validation of the BiSKit Sampler for the Detection of Microorganisms on Surfaces. <u>Biodefense Research Conference</u>, Abstr. 313, American Society for Microbiology, March 11, Baltimore, MD.

Session Organizer

<u>Fungal Biocontaminants in Indoor Environments</u>. American Society for Microbiology Colloquium, 103rd General Meeting of the American Society for Microbiology, May 20, Washington DC.

2002:

Invited Presentations

Fungal Contamination in Indoor Environments. <u>State of the Art</u>, National Institute of Disaster Restoration and Water Loss Institute Conference and Exposition, May 3, Chicago, IL.

Fungal Contamination in Indoor Environments. California Association for Medical Laboratory Technology Winter Seminar-North, March 23, Sacramento, CA.

Presentations

Bourget, S., P., Jacoby-Garrett, N. Burns-Savage, V. Stevens, P. Messier, and L. Stetzenbach. Effectiveness of Triosyn Filters in Reducing Airborne Microbial Concentrations in an Environmental Chamber. Abstr. Q7, Proceedings of the 102nd General Meeting of the American Society for Microbiology, May 20, Salt Lake City, UT.

Buttner, M.P., P. Cruz-Perez, **L.D. Stetzenbach**. Development of Biocontaminant Detection Strategies for Contaminated Surfaces. 2002 MASINT Biological Warfare Science and Technology Symposium, April 30-May 2, Monterey, CA.

Cruz, P., J.L. Henry, A.K. Klima-Comba, and L.D. Stetzenbach. Air and Surface Sampling of Fungal Contaminants in Indoor Environments. <u>Indoor Air 2002</u>, 9th International Conference on Indoor Air Quality and Climate, June 30-July 5, Monterey, CA.

Henry, J.L., P. Cruz, and **L.D. Stetzenbach**. Air and Surface Sampling in Residential Environments. Abstr. Q9, Proceedings of the 102nd General Meeting of the American Society for Microbiology, May 20, Salt Lake City, UT.

2001:

Invited Presentations

Biocontaminants in Indoor Air Quality. American Association of Bioanalysts Symposium, May 5, Las Vegas, NV.

Indoor Air Quality I - Microbial Contaminants of Indoor Environments. California Association of Public Health Directors Annual Meeting, April 24, Newport Beach, CA.

Indoor Air Quality II - Sampling and Analysis. California Association of Public Health Directors Annual Meeting, April 24, Newport Beach, CA.

Microbial Contamination. National Air Filtration Association Symposium, April 20, Las Vegas, NV.

Fungal Contaminants in Indoor Environments. University of Texas, Health Science Center at San Antonio, Teleconference Network of Texas, April 10.

Biological Airborne Particulate Matter. Kansas State University Symposium, January 8, Manhattan KS.

Presentations

Buttner, M.P., and L.D. Stetzenbach. Fungal Spores Dispersed from Fiberglass Ductboard, Fiberglass liner, and Galvanized Metal Air Handling System Duct Material. <u>Moisture, Microbes and Health Effects: Indoor Air Quality and Moisture in Buildings</u>, American Society of Heating, Refrigeration and Air Conditioning Engineers (ASHRAE) IAQ 2001 Conference, November 4-7, San Francisco, CA.

Buttner, M.P., P. Cruz, and **L.D. Stetzenbach**. Detection and Quantitation of *Aspergillus fumigatus* in Pure Culture using Polymerase Chain Reaction. Abstr. F-124, Proceedings of the 101th General Meeting of the American Society for Microbiology, May 23, Orlando, FL.

Cruz, P., M.P. Buttner and **L.D. Stetzenbach**. Specific Detection of *Stachybotrys chartarum* in Pure Culture using Polymerase Chain Reaction. Abstr. Q-145, Proceedings of the 101th General Meeting of the American Society for Microbiology, May 23, Orlando, FL.

Stetzenbach, L.D., M.P. Buttner, and A. Alavi. Evaluation of the Efficacy of Four Carpet Cleaning Methods in Reducing Fungal Contamination in Carpet. Abstr. Q-438, Proceedings of the 101th General Meeting of the American Society for Microbiology, May 23, Orlando, FL.

Buttner, M.P., P. Cruz, and **L.D. Stetzenbach**. Development of Biocontaminant Detection/Identification Strategies for CBrN Countermeasures. 2001 MASINT Biological Warfare Science and Technology Symposium, February 14, Elgin Air Force Base, FL.

2000:

Invited Presentations

Microbial Contamination in Indoor Environments. Texas Tech University Medical Center, October 5, Lubbock TX.

Microbiology of Bioaerosols. National Institute for Occupational Safety and Health (NIOSH), September 13-14, Morgantown, WV.

Microorganisms Indoor - are they making you ill? American Society for Clinical Laboratory Science Annual Meeting, July 26, San Francisco, CA.

Biocontaminants - how do you know if they are present? American Society for Clinical Laboratory Science Annual Meeting, July 26, San Francisco, CA.

Bioaerosols. <u>Waterborne Exposure Route for Microbial Gastrointestinal Illness and Other Diseases</u>, US Environmental Protection Agency Symposium, US Environmental Protection Agency, Office of Water, June 13-14, Arlington, VA.

Suspended in Time and Space. Divisional Group Symposium - <u>Connecting the Ecology of Pathogenic Microorganisms and Disease Outbreaks</u>, 100th General Meeting of the American Society for Microbiology, May 22, Los Angeles, CA.

Presentations

Jacoby-Garrett, P.M., and **L.D. Stetzenbach**. A Comparison of the AGI-30 and SKC BioSampler Air Samplers in Retrieval of Microbial Aerosols from a Controlled Environmental Chamber. Abstr. Q112, Proceedings of the 100th General Meeting of the American Society for Microbiology, May 23, Los Angeles, CA.

Henry, J. L., and **L.D. Stetzenbach**. Isolation of *Stachybotrys chartarum* from Environmental Samples using a Cellulose-based Medium. Abstr. Q114, Proceedings of the 100th General Meeting of the American Society for Microbiology, May 23, Los Angeles, CA.

Buttner, M.P., and **L.D. Stetzenbach.** Application of Molecular Methods for Enhanced Detection of Surface-associated Bacteria in Indoor Environments. 2000 MASINT Biological Warfare Science and Technology Symposium, January 13, Long Beach, CA.

Stetzenbach, L.D. Measurement of Biocontaminants Archived in Liquid Collection Buffers. 2000 MASINT Biological Warfare Science and Technology Symposium, January 13, Long Beach, CA.

1999:

Invited Presentations

Microorganisms and Indoor Air Quality. Indian Health Service symposium, sponsored by the Mid Atlantic Environmental Hygiene Resource Center, December 3, Albuquerque, MN.

Indoor Air Quality - Investigating and Resolving the Microbial Contamination of Indoor Air. National Environmental Health Association Conference, July 7, Nashville, TN. Indoor Air Quality - the Great *Stachybotrys* Debate. National Environmental Health Association Conference, July 8, Nashville, TN.

Analysis of Biological Detection Technologies. National Academy of Science. January 11-12, Washington, DC.

Presentations

Stetzenbach, L.D., M.P. Buttner, and P. Cruz-Perez. Dispersal of Fungal Spores from Contaminated Flooring Materials. The International Academy of Indoor Air Science (IAIAS), August 10, Edinburgh, Scotland.

Buttner, M.P., P. Cruz-Perez, P. Garrett, and L.D. Stetzenbach. Dispersal of Spores from Fungal-Contaminated Duct Material. The International Academy of Indoor Air Science (IAIAS), August 10, Edinburgh, Scotland.

Fungal Spores Aerosolized from Contaminated Flooring Materials. <u>Environmental Microbiology:</u> <u>Bioaerosols and Biosafety</u>, American Industrial Hygiene Conference and Exposition, June 9, Toronto, Canada.

Buttner, M.P., P. Cruz-Perez, P.J. Garrett, and L.D. Stetzenbach. A Comparison of the Airborne Dispersal of Fungal Spores from Contaminated Flooring Materials following Human Activity. Abstr. Q232, Proceedings of the 99th General Meeting of the American Society for Microbiology. June 1, Chicago, IL.

Buttner, M.P., P. Cruz-Perez, and **L.D. Stetzenbach**. Sampling Methods for the Detection and Quantitation of Surface-associated Microorganisms using Polymerase Chain Reaction. Abstr. Q229, Proceedings of the 99th General Meeting of the American Society for Microbiology, June 1, Chicago, IL.

Microorganisms and Indoor Air Quality. <u>Advances in Indoor Air Quality</u>. American Society for Microbiology Workshop, 99th General Meeting of the American Society for Microbiology, May 29, Chicago, IL.

Test Methods to Assess Contamination in HVAC Systems. Indoor Environment >99. April 21, Austin, TX.

Enhanced Detection of Airborne and Surface-associated Biocontaminants. American Association for the Advancement of Science Workshop - Research in Domestic Preparedness: Ensuring Security, Protecting Infrastructure, and Preventing Violence. March 15-16, Stillwater, OK.

1998:

Invited Presentations

National Aeronautics and Space Administration (NASA) strategic planning workshop - Environmental Health for Space Station and Mars Mission. December 7-9, Washington, DC.

Indoor Re-aerosolization. Joint Department of Defense and Department of Energy Urban Hazard Modeling workshop. August 25-26, Alexandria, VA.

Indoor Air Quality. Chemical Specialties Manufacturers = Association Indoor Air Quality Committee Meeting, May 28, Chicago, IL.

Olin Corporation Workshop on Indoor Air Quality, Drexel University, May 7, Philadelphia, PA

Monitoring and Measuring Contaminant Concentrations II Technical Session, Indoor Environment >98, April 15, Washington, DC

Naval Surface Warfare Center Information Exchange on Bio-aerosol Sample Collection, March 18, Dahlgren, VA.

Presentations

Diagnosing the Cause of a >Sick Building= - a Case Study of an Epidemiological and Microbiological Investigation. 3rd International Conference on Bioaerosols, Fungi, and Mycotoxins. September 23-25, Saratoga Springs, NY.

Indoor Air Quality. National Environmental Health Association workshop. 62nd Annual Education Conference and Exposition, June 28, Las Vegas, NV.

Fungal Spores Dispersed from Air Handling System duct - a Comparison of Three Fungal-Contaminated Duct Materials. American Industrial Hygiene Conference and Exposition, Indoor Air Ouality - Session II, May 13, Atlanta, GA.

Microorganisms and Indoor Air Quality. Indoor Air Quality. American Society for Microbiology Workshop, 98th General meeting of the American Society for Microbiology, May 17, Atlanta, GA

Microbiological Contaminants of Indoor Environments. Microbiological Contaminants of Indoor Environments, presented by the Mid-Atlantic Environmental Hygiene Resource Center, PA, February 4-6 New Orleans, LA.

Sampling Techniques and Procedures for Microbiological Contaminants. <u>Microbiological Contaminants of Indoor Environments</u>, presented by the Mid-Atlantic Environmental Hygiene Resource Center, PA, February 4-6 New Orleans, LA.

Session Organizer

Indoor Air Quality. American Society for Microbiology Workshop. 98th General Meeting of the American Society for Microbiology, May 17, Atlanta, GA.

1997: Invited Presentations Microorganisms and Indoor Air Quality. <u>Indoor Air Quality</u>, PDA Training and Research Institute Workshop, PDA Training and Research Institute, November 14, Philadelphia, PA.

Microorganisms and Indoor Air Quality. <u>A New Age for Microbiology</u>, American Society for Microbiology Southern California Branch Annual Meeting, November 8, Irvine, CA.

Microbes and the Indoor Air Environment. <u>Microorganisms and Indoor Environmental Quality</u>. Society for Industrial Microbiology Annual Meeting, August 5, Reno, NV. Microorganisms and Indoor Air Quality. <u>Indoor Air Quality</u>. American Society for Microbiology Workshop. 97th General Meeting of the American Society for Microbiology, May 4, Miami Beach, FL.

Presentations

Dispersal of fungal spores from Contaminated Metal and Fiberglass Duct. <u>Engineering Solutions to Indoor Air Quality Problems</u> symposium, Air and Waste Management Assoc. and the US Environmental Protection Agency, July 21-23, Research Triangle Park, NC.

Microbiological contaminants of indoor environments: bacteria, virus, and other microorganisms. Biological Contaminants of Indoor Environments, Mid-Atlantic Environmental Hygiene Resource Center, PA, US Environmental Protection Agency, Region 5, and the National Association of County and City Health Officials, April 16-18, Chicago, IL.

Sampling and Analysis Techniques and Procedures. <u>Biological Contaminants of Indoor Environments</u>, Mid-Atlantic Environmental Hygiene Resource Center, PA, US Environmental Protection Agency, Region 5, and the National Association of County and City Health Officials, April 16-18, Chicago, IL.

Session Organizer

Indoor Air Quality. PDA Training and Research Institute Workshop, PDA Training and Research Institute. November 14, Philadelphia, PA.

<u>Microorganisms and Indoor Environmental Quality</u>. Society for Industrial Microbiology Symposium, Society for Industrial Microbiology Annual Meeting, August 5, Reno, NV.

<u>Indoor Air Quality</u>. American Society for Microbiology symposium. 97th General Meeting of the American Society for Microbiology, May 4, Miami Beach, FL.

1996:

Invited Presentations

Indoor Air Quality - Microbiological Considerations. 5th Western Pacific Congress of Chemotherapy and Infectious Diseases, December 1, Singapore.

Carlyn Halde Foundation Lecturer: Environmental Mycology - *Stachybotrys atra* Case Studies. American Society for Microbiology Northern California Branch and the Northern California Association of Public Health Microbiologists, November 15, San Jose, CA.

Microorganisms and Indoor Air Quality. <u>Indoor Air Quality</u>. American Society for Microbiology Workshop, 96th General Meeting of the American Society for Microbiology, May 19, New Orleans, LA.

Presentations

Buttner, M.P., L.E.M. Cole, P. Cruz-Perez, and **L.D. Stetzenbach**. Comparison of Selected Culture Media for the Retrieval of Airborne Fungi. Abstr. Q11, Proceedings of the 96th General Meeting of the American Society for Microbiology.

Dvorsky, E., G.A.Toranzos and **L.D. Stetzenbach**. An Alternative Method for the Detection of Airborne *Legionella* spp. Abstr. Q17, Proceedings of the 96th General Meeting of the American Society for Microbiology.

Microbiological Contaminants of Indoor Environments - Bacteria, Virus, and Other Microorganisms. Biological Contaminants of Indoor Environments, sponsored by the Mid-Atlantic Environmental Hygiene Resource Center, Philadelphia, PA, US Environmental Protection Agency, Region 9, and the University of Tulsa, March 27-29, San Diego, CA.

Sampling and Analysis Techniques and Procedures. <u>Biological Contaminants of Indoor Environments</u>, sponsored by the Mid-Atlantic Environmental Hygiene Resource Center, Philadelphia, PA, US Environmental Protection Agency, Region 9, and the University of Tulsa, March 27-29, San Diego, CA.

Session Organizer

<u>Indoor Air Quality</u>. American Society for Microbiology Workshop. 96th General Meeting of the American Society for Microbiology, May 19, New Orleans, LA.

1995:

Invited Presentations

Microorganisms and Indoor Air Quality. <u>Indoor Air Quality: Microbiological Considerations</u>. American Society for Microbiology workshop. 95th General Meeting of the American Society for Microbiology, May 21, Washington, DC.

Case studies. <u>Indoor Air Quality: Microbiological Considerations</u>. American Society for Microbiology Workshop. 95th General Meeting of the American Society for Microbiology, May 21, Washington, DC.

Presentations

Alvarez, A.J., M.P. Buttner, and **L.D. Stetzenbach.** Effects of Environmental Interference and Sampling Stress on PCR for Bioaerosol Monitoring. Abstr. Q368, Proceedings of the 95th General Meeting of the American Society for Microbiology, p. 464.

Microbiological Contaminants of Indoor Environments. <u>Biological Contaminants of Indoor Environments</u>. Mid-Atlantic Environmental Hygiene Resource Center Workshop, Philadelphia, PA, January 11-13, St. Petersburg, FL.

Sampling Techniques. <u>Biological Contaminants of Indoor Environments</u>. Mid-Atlantic Environmental Hygiene Resource Center Workshop, Philadelphia, PA, January 11-13, St. Petersburg, FL.

Session Organizer

<u>Indoor Air Quality: Microbiological Considerations</u>. American Society for Microbiology workshop. 95th General Meeting of the American Society for Microbiology, May 21, Washington, DC.

1994:

Invited Presentations

Microbial Indoor Air Quality. Keynote Speaker, Sociedad De Microbiologos de Puerto Rico, January 28, San Juan, Puerto Rico.

Microorganisms and Indoor Air Quality. <u>Indoor Air Quality</u>, <u>Part I</u>. American Society for Microbiology workshop. 94th General Meeting of the American Society for Microbiology, May 22-23, Las Vegas, NV.

Identification of Airborne Bacteria. <u>Indoor Air Quality, Part II</u>. American Society for Microbiology Workshop. 94th General Meeting of the American Society for Microbiology, May 22-23, Las Vegas, NV.

Microbial Aerosols. <u>Methods for Detecting Microorganisms in the Environment: What's New?</u> What's Next? American Society for Microbiology Symposium. 94th General Meeting of the American Society for Microbiology, May 26, Las Vegas, NV.

Presentations

<u>Progress in Methods Development for Monitoring Microorganisms in Bioaerosols</u>, American Society for Microbiology Symposium Roundtable. 94th General Meeting of the American Society for Microbiology, May 27, Las Vegas, NV.

The Microbiology of Indoor Environments. <u>Assessing Microbiological Contamination of Indoor Environments</u> symposium. Mid-Atlantic Environmental Hygiene Resource Center, Philadelphia, PA, April 6-8, Fairfax, VA.

Monitoring for Airborne Bioaerosols. <u>Assessing Microbiological Contamination of Indoor Environments</u>. Mid-Atlantic Environmental Hygiene Resource Center, Philadelphia, PA, April 6-8, Fairfax, VA.

Session Organizer

<u>Indoor Air Quality, Part I and Part II</u>. American Society for Microbiology workshop. 94th General Meeting of the American Society for Microbiology, May 22-23, Las Vegas, NV.

Methods for Detecting Microorganisms in the Environment: What's New? What's Next? American Society for Microbiology Symposium, 94th General Meeting of the American Society for Microbiology, May 26, Las Vegas, NV.

Honors/Appointments/Committees

Editor, 2001-2004. Applied and Environmental Microbiology, American Society for Microbiology, Washington, DC.

Editorial Board, 2004-2006. Manual of Environmental Microbiology, 3rd edition in preparation, ASM Press, Washington, DC.

Editorial Board, 2002. Encyclopedia of Environmental Microbiology, John Wiley and Sons, Inc., New York.

Editorial Board, 2002, Manual of Environmental Microbiology, 2nd edition, ASM Press, Washington, DC.

Editorial Board, 2000-2002. Microbial Ecology, Springer, New York.

Editorial Board, 1999-2001. Aerobiologia, Kluwer Academic Publishers, The Netherlands.

Editorial Board, 1999-2001. Journal of Industrial Microbiology and Biotechnology, Stockton Press, England.

Editorial Board, 1995-1997 and 1998-2000. Applied and Environmental Microbiology, American Society for Microbiology, Washington, DC.

Editorial Board, 1997, Manual of Environmental Microbiology, ASM Press, Washington, DC.

Committee Chair, Institutional Biosafety Committee, 2005 – present. University of Nevada, Las Vegas.

Member, Waksman Foundation for Microbiology Lectures Program, 2005-2007. American Society for Microbiology, Washington, DC.

Member, Science Advisory Board, Homeland Security Advisory Committee, 2005 – present. Office of the Administrator, US Environmental Protection Agency, Washington, DC.

Member, Committee on Environmental Microbiology, 2001-2007. Public and Scientific Affairs Board, American Society for Microbiology, Washington, DC.

Member, Board of Directors, 2004-present. National Center for Energy Management and Building Technologies, Alexandria, VA.

External Advisory Board Member, 2001-2006. Cincinnati Childhood Allergy and Air Pollution Study, University of Cincinnati, Cincinnati, OH.

Member, Advisory Board, 2002. Defense Threat Reduction Agency, Ft. Belvoir, VA.

Member, Special Project Committee (SPC 180P – Standard Practice for Inspection and Maintenance for HVAC Systems), 2004-present. American Society of Heating, Refrigeration, and Air Conditioning Engineers (ASHRAE), Atlanta, GA.

Member, Bioaerosols Committee, American Conference of Governmental Industrial Hygienists (ACGIH), 2004-present.

Member, Environmental Health Committee, 2001-2004. American Society of Heating, Refrigeration, and Air Conditioning Engineers (ASHRAE), Atlanta, GA.

Peer Review Panelist, July 2004. National Institutes of Health ZRG1 IDM-B 12 V.E.P. Special Emphasis Panel, Washington DC.

Peer Review Panelist, March 2004. National Institutes of Health ZRG1 SSS-Z 10B Study Section, Washington DC.

Peer Review Panelist, November 2003. National Institutes of Health, Washington, DC.

Peer Review Panelist, May 2003. National Institutes of Allergy and Infectious Diseases/National Institutes of Health, Planning Grants for Regional Centers of Excellence for Bioterrorism and Emerging Infectious Diseases, Gaithersburg, MD.

Peer Review Panelist, March 2003. National Institutes of Health, Washington, DC.

Peer Review Panelist, November 2002. National Institutes of Health SBIR/STTR Infectious Diseases and Microbiology Study Section, Washington, DC.

Peer Review Panelist, October 2002. Bacteriology and Mycology Subcommittee, National Institutes of Health, Washington, DC.

Study Panelist, January 11-12, 1999. National Academy of Science, Washington, DC.

Discussant, January 11-12. Workshop on the Methodology for Implementing Timely Incident Response, Water Environment Research Foundation, Alexandria, VA.

Chair, US Environmental Protection Agency, National Environmental Research Laboratory Peer Review Panel, Cincinnati, OH, July 2001.

Chair-Elect, 2001-2002 and Chair, 2002-2003. Environmental and General Applied Microbiologists - Division Q, American Society for Microbiology.

Alternate Councilor, 1992-1994 and Councilor, 1994-1996. Environmental and General Applied Microbiologists - Division Q, American Society for Microbiology.

Award of Excellence, March 1990. Environmental Monitoring Systems Laboratory - Las Vegas, U.S. Environmental Protection Agency. Recognition for participation in the US EPA bioremediation research project in Valdez, Alaska following the Exxon Valdez oil spill in Prince William Sound and in the US EPA Headquarters' indoor air quality monitoring survey.

Book Reviewer, 1995. Health Implications of Fungi in Indoor Environments, R.A.Samson, B. Flannigan, M.E. Flannigan, A.P. Verhoff, O.C.G. Adan, and E.S. Hoekstra (ed.), Elsevier Science, Inc., New York. Review published in Mycopathologia, Vol. 4, p. 1.

Professional Organizations

American Conference of Governmental Industrial Hygienists (ACGIH) (2004-present)

American Industrial Hygiene Association (1994-present)

American Society of Heating, Refrigerating, and Air Conditioning Engineers

(ASHRAE; 2002-present)

American Society for Microbiology (1983-present)

Indoor Air Quality Council, Las Vegas Chapter (2000-2004)

International Society of Indoor Air Quality and Climate (ISIAQ; 1999-present)

Mycological Society of America (1994-2004)

Nevada Public Health Association (2005 – present)

Research Funding

Governmental:

Lawrence Livermore National Laboratory (LLNL)

National Aeronautics and Space Administration (NASA)

National Institute of Occupational Safety and Health (NIOSH)

State of Nevada, Risk Management

U.S. Department of Defense

U.S. Department of Energy

U.S. Environmental Protection Agency

Industry:

E.I. DuPont de Nemours, Inc.

Exponent

Harris Research, Inc.

Masco Corporation

North American Insulation Manufacturers Association

S.C. Johnson and Sons, Inc.

Triosyn Corporation

Triton Systems

Graduate Faculty/Graduate Student Thesis/Dissertation Committees/Mentoring

Graduate Faculty and Course Organizer, Science of Catastrophic Incidents (ECEM 722), Executive Masters in Crisis and Emergency Management, University of Nevada, Las Vegas.

Associate Graduate Faculty, Department of Environmental Sciences and Health, University of Nevada, Reno.

Associate Graduate Faculty, Department of Environmental Sciences, University of Nevada, Las Vegas.

Associate Graduate Faculty, Department of Chemistry, University of Nevada, Las Vegas. 1995-2003.

Associate Graduate Faculty, Department of Health Physics, University for Nevada, Las Vegas. 1997-1999

Committee Member for Lazaro Eleuterio, 2005-present, Department of Civil and Environmental Engineering, University of Nevada, Las Vegas - Jacimaria Batista, Advisor.

Committee Member for Alex Aguiar, 2003-present, Department of Civil and Environmental Engineering, University of Nevada, Las Vegas - Jacimaria Batista, Advisor.

Committee Member for Shar Todd, Ph.D., degree awarded May 2003, University of Nevada, Reno-Stan Omaye, Advisor.

Dissertation Advisor to Patricia Cruz-Perez, Ph.D., degree awarded December 2000. Environmental Sciences and Health, University of Nevada, Reno.

Dissertation Advisor to Mark P. Buttner, Ph.D., degree awarded May 2000. Environmental Sciences and Health, University of Nevada, Reno.

Mentor for A.J. Alvarez, Ph.D. 1994-1995. National Science Foundation Minority Postdoctoral Research Fellowship Program.

Committee Member for Sandra Story, M.S. 1994. Department of Biological Sciences, University of Nevada, Las Vegas - Penny Amy, Advisor.

Patents/Licenses Awarded

University of Arizona (UA 805 - AMonoclonal Antibodies Unique to *Giardia* Cyst Wall Determinants@) licensed to Meridian Diagnostics, 1987. Co-inventor: Charles R. Sterling, Department of Veterinary Science, University of Arizona.

RELATED PROCEEDINGS APPENDIX

None